

A rapid screening procedure utilizing iododeoxyuridine-incorporated artificial **chromosomes** facilitated the assessment of different transfection conditions. The results were confirmed by cytogenetic analysis of positively transfected clones. By transfecting both hamster lung fibroblast cells (V79-4) and murine connective tissue cells [L-M(TK-)], the best results were obtained using either Superfect (cationic dendrimer) or LipofectAMINE 2000 (cationic lipid) with protocols adapted for metaphase **chromosome** preparation. Transfection efficiencies of 10^{-4} - 10^{-2} (0.01-1%) were routinely observed, and recipient cells were able to maintain expression of the reporter gene over the total length of the experiment. This represents a significant advance over our previous attempts at mass-transfection of artificial **chromosomes** using microcell fusion, where we routinely achieved efficiencies at least two orders of magnitudes less than reported here. These data are particularly noteworthy given that lipid-mediated gene transfer typically involves transfecting millions of plasmids (1 microg of DNA from a 5 kb plasmid is approximately 1.2×10^{11} copies) to each cell whereas the much larger artificial **chromosomes** comprise only a one-to-one ratio, yet achieve transfection efficiencies of (10^{-2} - 10^{-1}), that is, comparable to our results. These data suggest that artificial **chromosomes** containing therapeutic genes can be successfully delivered to target cells in vitro using well-established transfection agents.

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L1      16663 S ARTIFICIAL CHROMOSOM?
L2      23 S L1 AND LIPOFECTAMINE
L3      14 DUP REM L2 (9 DUPLICATES REMOVED)
L4      3 S L1 AND DOSPA AND DOPE
L5      78518 S LARGE (S) (NUCLEIC ACID OR DNA)
L6      37 S L5 AND LIPOFECTAMINE
L7      31 DUP REM L6 (6 DUPLICATES REMOVED)
L8      60 S CHROMOSOM? AND LIPOFECTAMINE
L9      34 DUP REM L8 (26 DUPLICATES REMOVED)
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Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	2	("6025155").PN.	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	OFF	2005/03/29 14:01
L2	259	dospa same dope	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:01
L3	0	l2 same "artificial chromosome"	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:02
L4	32	l2 and "artificial chromosome"	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:04
L5	17	lipofectamine same chromosome	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:05
L6	3036	lipofectamine and chromosome	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:06
L8	68	lipofectamine and ("artificial chromosome" same mammalian)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:10
L9	12	lipofectamine same aces	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/03/29 14:07